Supporting Information

Matlack et al. 10.1073/pnas.1402228111

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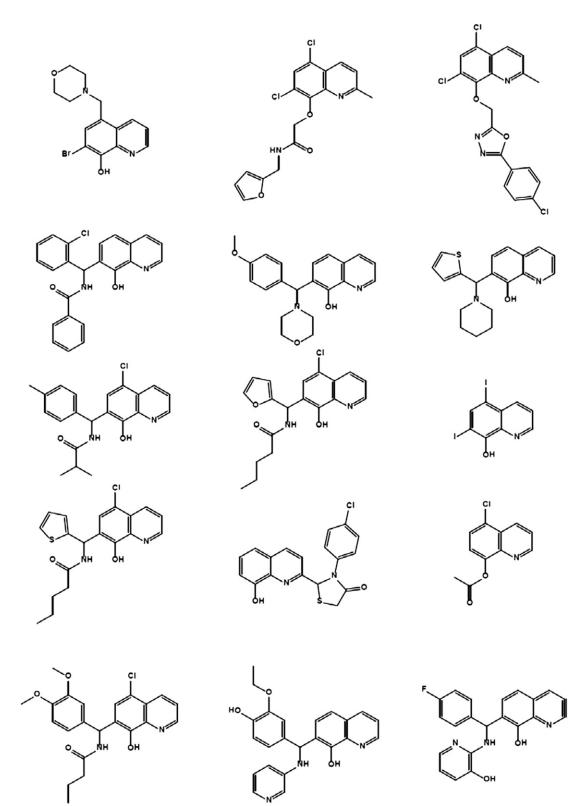


Fig. S1. Structures of 8-hydroxyquinoline (8-OHQ) hits. Fifteen of 30 hits from the initial β -amyloid (A β) small-molecule screen were 8-OHQs. All compounds had the core 8-OHQ with various, diverse substituents, largely attached through the hydroxyl-containing ring.

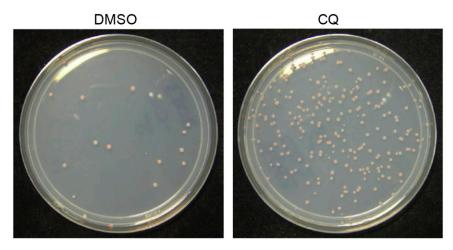


Fig. S2. Clioquinol (CQ) increases cell number. To ensure that the increase in OD₆₀₀ was due to a rescue of toxicity and not due to an artifact of increased cell size, cells were plated to single colony-forming units after CQ treatment. Indeed, there was an increase in the number of viable cells after treating A β -expressing cells with a protective dose of CQ.

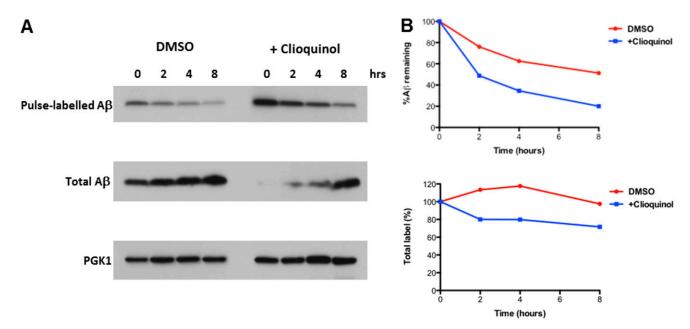


Fig. S3. ³⁵S pulse labeling of $A\beta$. (A) $A\beta$ analysis of pulse-labeling experiment. The top panel shows ³⁵S-labeled, immunoprecipitated $A\beta$ over time. The middle shows total $A\beta$ by immunoblot with the 6E10 antibody. The bottom panel shows levels of control protein, Pgk1. (*B*) Quantification of $A\beta$ and total ³⁵S labeling during degradation time course. The top panel is exact data shown in Fig. 6. The bottom panel shows total label as quantified from an entire lane of protein without immunoprecipitation.

DNA C

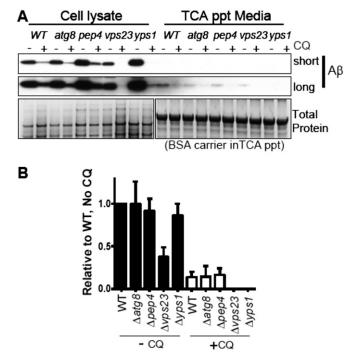


Fig. 54. Genetic ablation of major degradation pathways does not reduce $A\beta$ levels in response to CQ. (*A*) Immunoblot analysis of $A\beta$ from cell lysates and media after treating $A\beta$ strains with deletions of different degradation pathways. Media was also trichloroacetic acid (TCA) precipitated to monitor whether $A\beta$ was released into the media upon CQ treatment. BSA was added to the media just before TCA precipitation to ensure quantitative protein precipitation. This is observed in the "Total Protein" Coomassie-stained gel. Genes deleted were *ATG8* (autophagy), *PEP4* (vacuolar protease), *VPS23* (endosome to vacuole), and *YPS1* (major aspartic protease localized to plasma membrane). (*B*) Quantitation of *A*. Secondary antibodies were IRDye infrared dye-linked antibodies scanned with the Li-Cor Odyssey Scanner. The $\Delta vps23$ strain routinely had lower $A\beta$ levels. Both $\Delta vps23$ and $\Delta yps1$ typically resulted in an even more complete degradation of $A\beta$ peptide in response to CQ.

Table S1. Yeast strains

Strain	Genotype	
Αβ	MATα, can1-100, his3-11,15, leu2-3,112, <i>TRP1</i> ::ssAβ, <i>URA3</i> ::ssAβ, ade2-1 <i>pdr5</i> ::KAN	This study
YFP	MATα, can1-100, his3-11,15, leu2-3,112, <i>TRP1</i> ::YFP, <i>URA3</i> ::YFP, ade2-1 <i>pdr5</i> ::KAN	Ref. 1
Aβ MUP1-GFP	ΜΑΤα, can1-100, his3-11,15, leu2-3,112, <i>TRP1</i> ::ssAβ, URA3::ssAβ, ade2-1 <i>pdr5</i> ::KAN <i>MUP1-GFP::HIS3</i>	This study
MUP1-GFP	MAΤα, can1-100, his3-11,15, leu2-3,112, ade2-1 <i>pdr1::KAN pdr3::KAN</i>	Ref. 2
Aβ <i>∆atg8</i>	ΜΑΤα, can1-100, his3-11,15, leu2-3,112, <i>TRP1</i> ::ssAβ, <i>URA3</i> ::ssAβ, ade2-1 <i>pdr5</i> ::KAN, <i>atg8</i> ::HPH	This study
Αβ Δρερ4	ΜΑΤα, can1-100, his3-11,15, leu2-3,112, <i>TRP1</i> ::ssAβ, <i>URA3</i> ::ssAβ, ade2-1 <i>pdr5</i> ::KAN, <i>pep4:</i> :HPH	This study
Aβ Δvps23	ΜΑΤα, can1-100, his3-11,15, leu2-3,112, <i>TRP1</i> ::ssAβ, <i>URA3</i> ::ssAβ, ade2-1 <i>pdr5</i> ::KAN, <i>vps23</i> ::HPH	This study
Αβ Δyps1	MATα, can1-100, his3-11,15, leu2-3,112, <i>TRP1</i> ::ssAβ, <i>URA3</i> ::ssAβ, ade2-1 <i>pdr5</i> ::KAN, <i>yps1:</i> :HPH	This study

1. Su LJ, et al. (2010) Compounds from an unbiased chemical screen reverse both ER-to-Golgi trafficking defects and mitochondrial dysfunction in Parkinson's disease models. Dis Model Mech 3(3-4):194–208.

2. Tardiff DF, et al. (2013) Yeast reveal a "druggable" Rsp5/Nedd4 network that ameliorates α-synuclein toxicity in neurons. Science 342(6161):979–983.

Table S2.	Gene deletions and	l toxic compound	tested for rescue b	y CQ
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Yeast gene deletion/drug	Gene name	Mechanism	Significant rescue by CQ?
Systematic yeast name			
YEL046C	GLY1		No
YGL070C	RPB9		No
YCR028C	FEN2		No
YDR463W	BAP1		No
YGR167W	CLC1		No
Drug			
Trifluoperazine		H+ ATPase, K ⁺ regulation	No
t-Butyl hydrogen peroxide		Oxidant	No
Rapamycin		TOR inhibition	Mild
4-Nitroquinoline-1-oxide		DNA damage	No
Cycloheximide		Translation inhibitor	No
Gliotoxin		Thioredoxin redox	No
Hydroxyurea		dNTP depletion	No

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